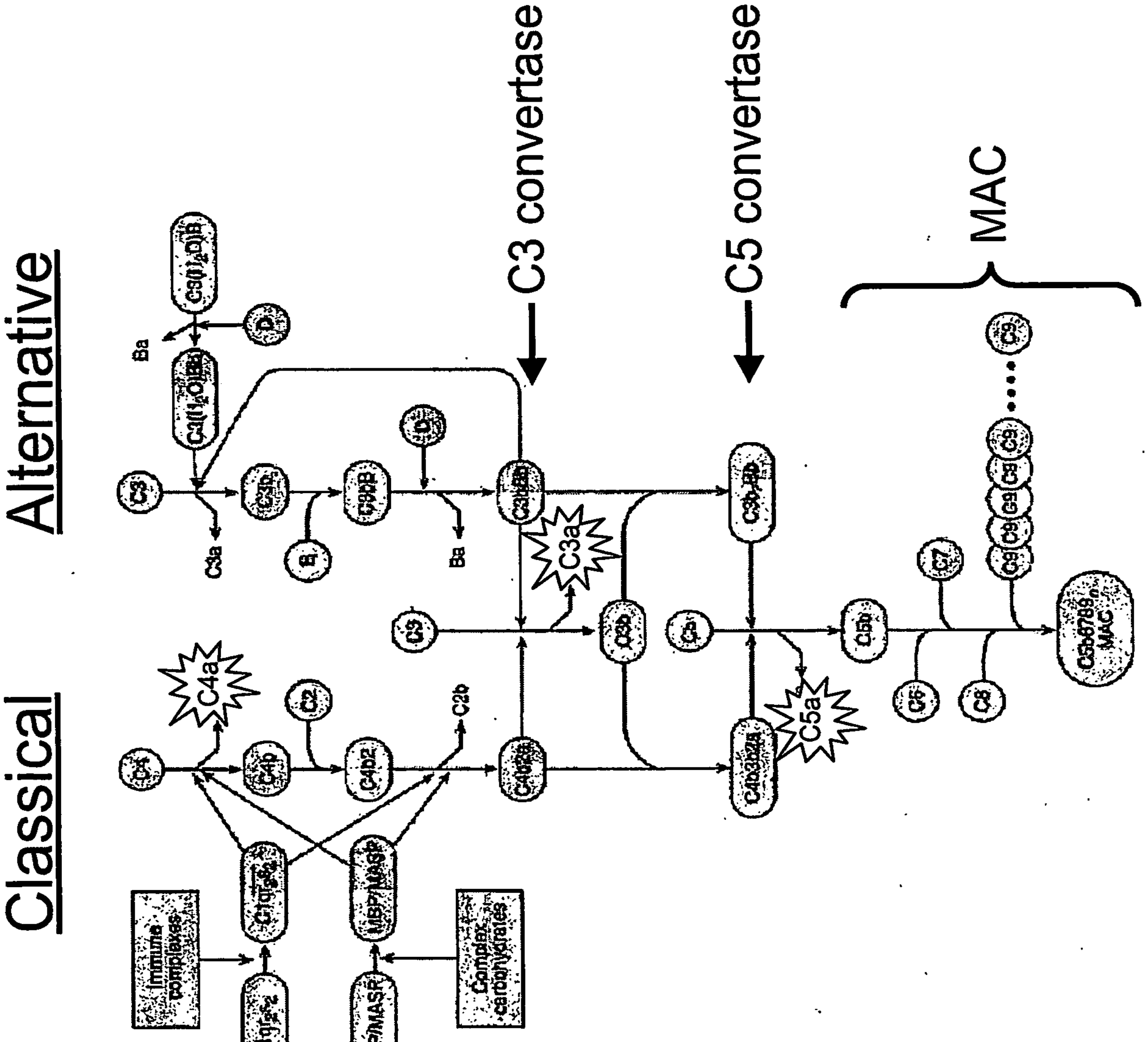

Classical
Alternative
C3 convertase
C5 convertase
MAC
Immune complexes
Complex carbohydrates
C1q1r2s2
C1q1r2s2
MBP/MASP
MBP/MASP
C4
C4a
C4b
C2
C4b2
C2b
C4b2a
C3
C3a
C3b
C4b3b2a
C5
C5a
C5b
C6
C7
C8
C9
C9
C9
C9
C9
C9
C5b6789n MAC
C3
C3a
C3b
B
C3bB
D
Ba
C3bBb
C3b2Bb
C3(H2O)
C3(H2O)B
C3(H2O)Bb
D
Ba

Fig. 1

Fig. 2

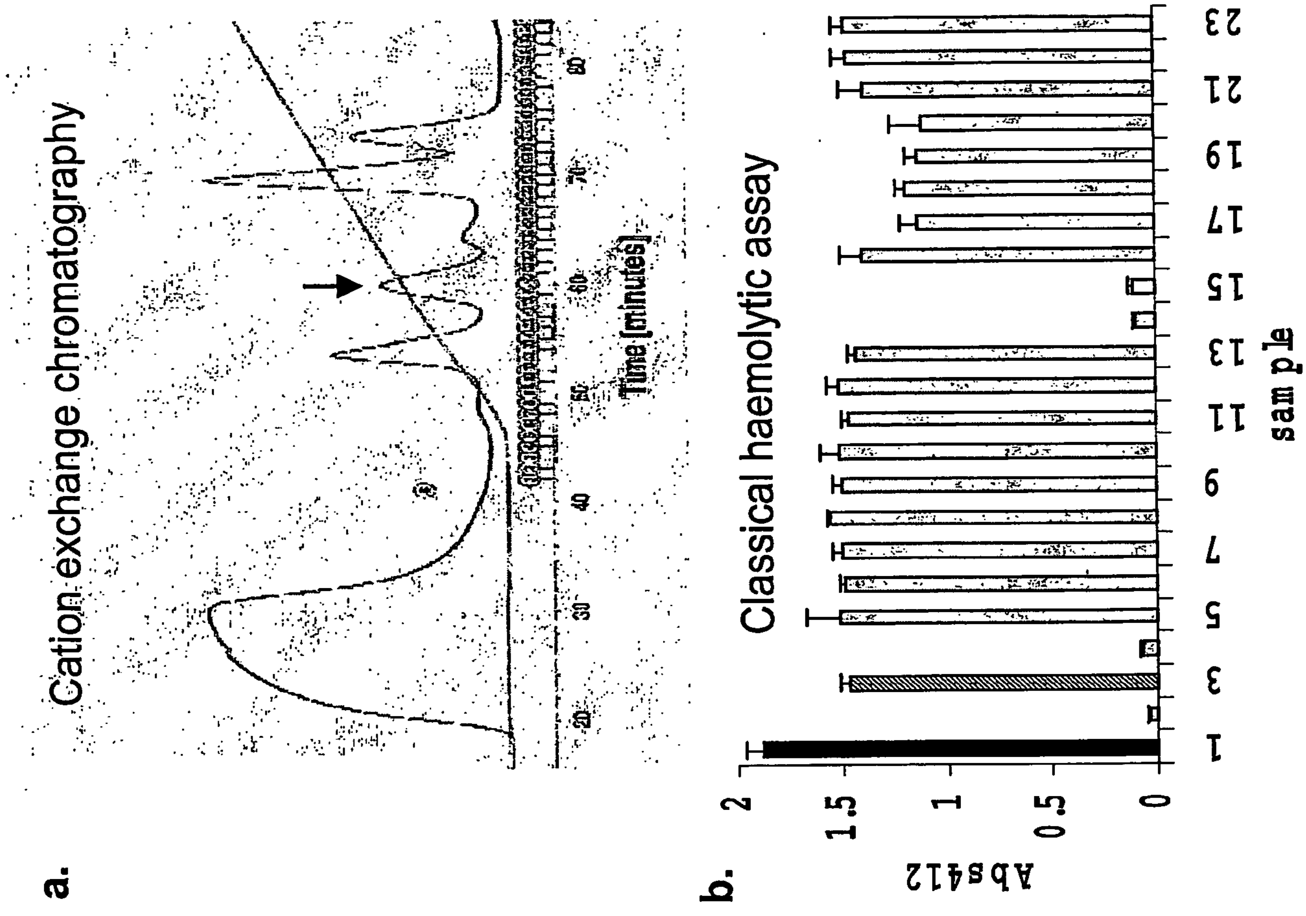

a.
Cation exchange chromatography
20
30
40
50
60
70
80
Time [minutes]
b.
Classical haemolytic assay
Abs412
2
1.5
1
0.5
0
1
3
5
7
9
11
13
15
17
19
21
23
sample

Fig. 3

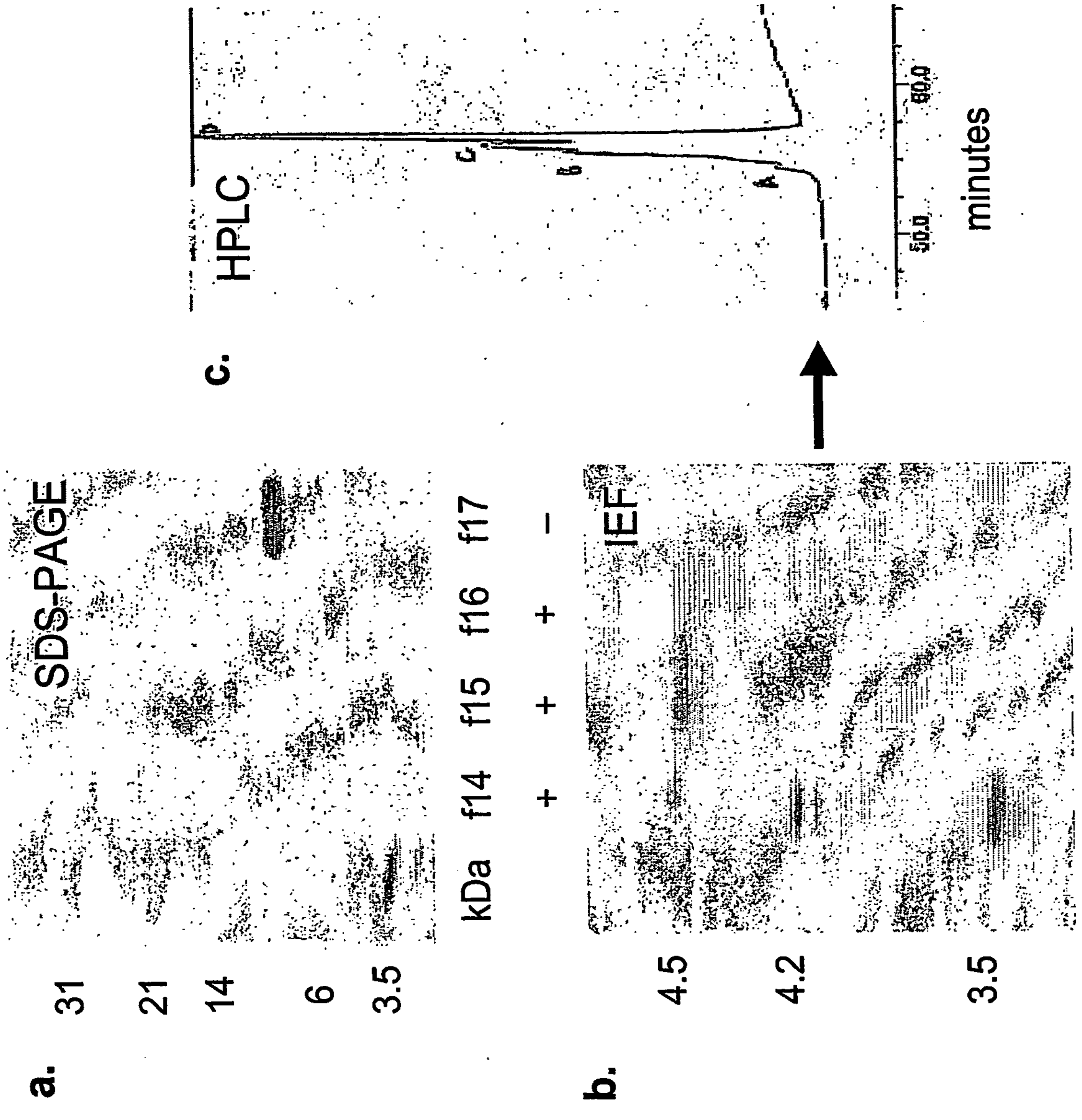

a.
SDS-PAGE
kDa
31
21
14
6
3.5
f14
f15
f16
f17
+
+
+
–
b.
IEF
PI
4.5
4.2
3.5
f17
f15
–
+
c.
HPLC
A
B
C
50.0
60.0
minutes

Fig. 4

ATGCTGGTTTTGGTGACCCTGATTTTCTCCTTTTCTGCGAACATCGCATATGCTGACAGC 60
M L V L V T L I F S F S A N I A Y A D S 20

GAAAGCGACTGCACTGGAAGCGAACCTGTTGACGCCTTCCAAGCTTTCAGTGAGGGCAAA 120
E S D **C** T G S E P V D A F Q A F S E G K 40

GAGGCATATGTCCTGGTGAGGTCCACGGATCCCAAAGCGAGGGACTGCTTGAAAGGAGAA 180
E A Y V L V R S T D P K A R D **C** L K G E 60

CCAGCCGGAGAAAAGCAGGACAACACGTTGCCGGTGATGATGACGTTTAAGAATGGCACA 240
P A G E K Q D N T L P V M M T F K N G T 80

GACTGGGCTTCAACCGATTGGACGTTTACTTTGGACGGCGCAAAGGTAACGGCAACCCTT 300
D W A S T D W T F T L D G A K V T A T L 100

GGTAACCTAACCCAAAATAGGGAAGTGGTCTACGACTCGCAAAGTCATCACTGCCACGTT 360
G N L T Q N R E V V Y D S Q S H H **C** H V 120

GACAAGGTCGAGAAGGAAGTTCCAGATTATGAGATGTGGATGCTCGATGCGGGAGGGCTT 420
D K V E K E V P D Y E M W M L D A G G L 140

GAAGTGGAAGTCGAGTGCTGCCGTCAAAAGCTTGAAGAGTTGGCGTCTGGCAGGAACCAA 480
E V E V E **C** **C** R Q K L E E L A S G R N Q 160

ATGTATCCCCATCTCAAGGACTGCTAG 507
M Y P H L K D **C** * 168

Fig. 5

```
OmCI      .MLVLVTLIFSESANIAYADSESDCSGSEPVDAFQAFSEGKEAYVLVRST
TSGP3     MMLVLATVILSFSASTALAD....CPTGKPTEAYVAFNEGKGAYILVRST
TSGP2     MMLVLATVILSFSASTALAD....CPTGKPTDAYVAFNEGQGAYILVKST
Moubatin  MMLVLTTLIFSFSASIAYAQSG..CSVSDPLDALKAFKDGAGTFLLQKST
           **** * * ****  * *     *    *  *  **  *     *  **

OmCI      DPKARDCLKGEPAGEKQDNTLPVMMTFKNGTDWASTDWTETLDGAKVTAT
TSGP3     NLNARDCLKGEATGKKEGNTLPVMMAFKDEGKWVSLPWTFTLDGPKVTAT
TSGP2     DLDARDCLKGSATGKKEGNKVPVMMAFKNEGQWVSLPWTFTLDGPKVTAT
Moubatin  DPQARDCLKGTPNGNRDGNTLPVTMTYKDDSKWVSLNWMFTLEGANIVAT
             *******   *    *  ** *  *    * *  * *** *    **

OmCI      L.GNLTQNREVVYDSQSHHCHVDKVEKEVPDYEMWMLDAGGLEVEVECCR
TSGP3     H.GQRTLKGEVVYDVPSHHCHIEKLESGA..YDMWMLEAGGLEVDIECCN
TSGP2     D.GQRTLKREVVYDVASHHCHVEKLASGA..YEMWMLEAGGLEVDIECCN
Moubatin  LEGKRKQRGELVYDVQSHDCHITKLSSGV..YQQWQSNGSADDKDIKCCD
            *      * ***   * **  *       *  *            **

OmCI      QKLEELASGRNQMYPHLKDC.....
TSGP3     KRYDELTSGQVVIRPQDKDC.....
TSGP2     KKYDELTSGQVVIRPQDKDC.....
Moubatin  EKFKELTSGIDYTKPQEKGCETSAK
              ** **     *  * *
```

Fig. 6

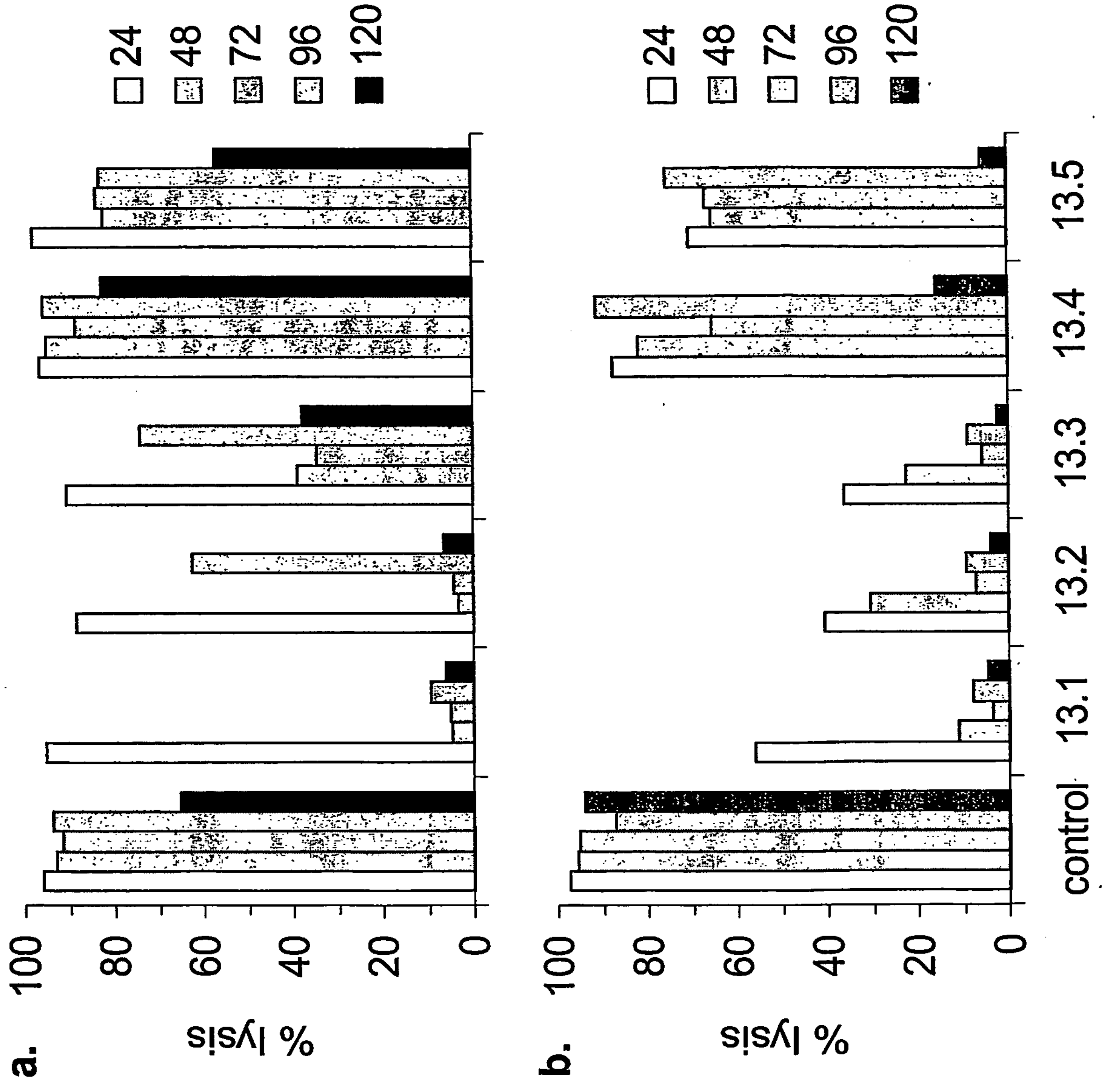
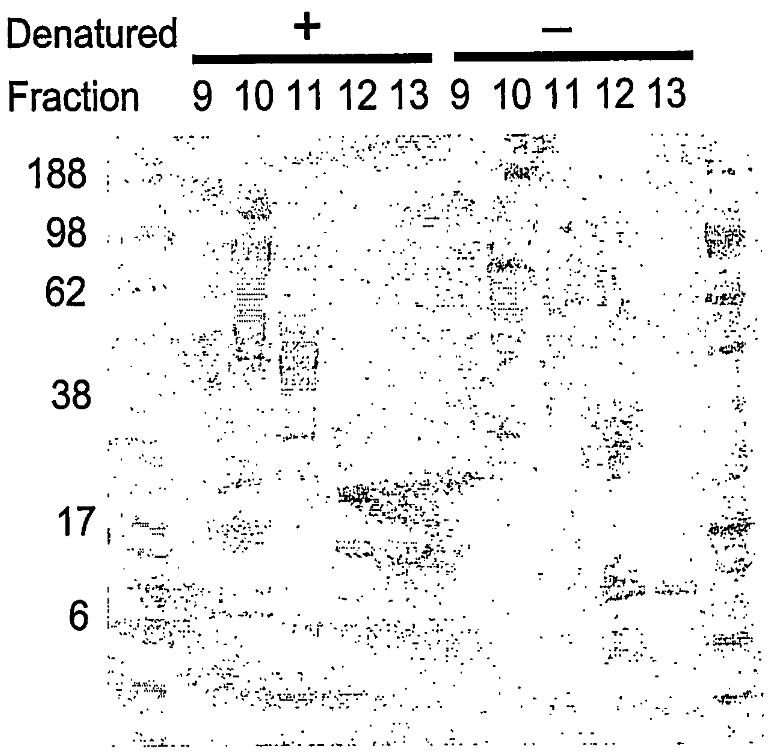

a.
% lysis
100
80
60
40
20
0
control
13.1
13.2
13.3
13.4
13.5
□ 24
▨ 48
▨ 72
▨ 96
■ 120
b.
% lysis
100
80
60
40
20
0
□ 24
▨ 48
□ 72
▨ 96
■ 120

Fig. 7a


Denatured
+
–
Fraction
9 10 11 12 13 9 10 11 12 13
188
98
62
38
17
6

b

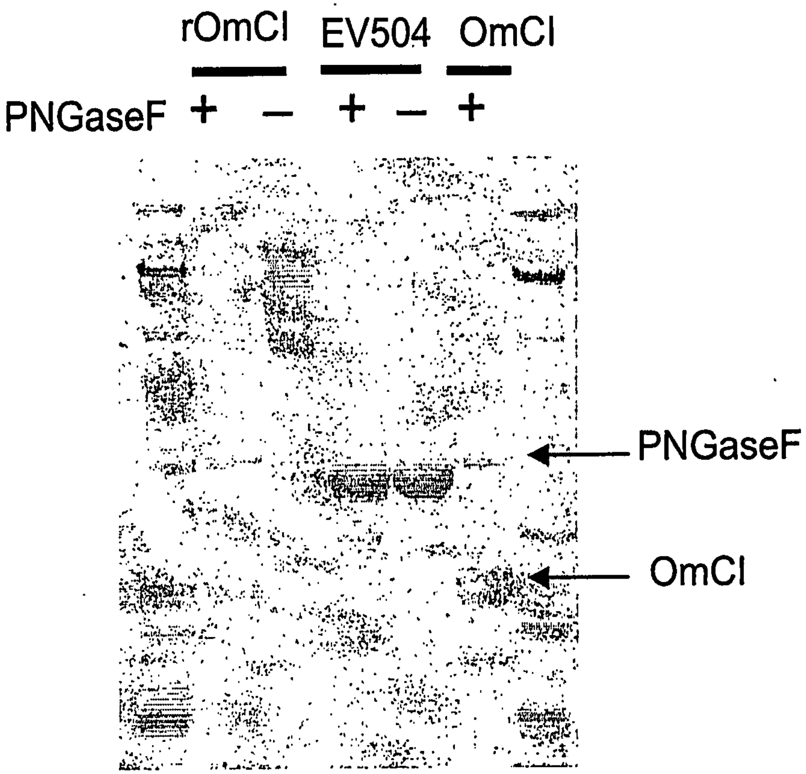

rOmCI
EV504
OmCI
PNGaseF
+ – + – +
PNGaseF
OmCI

Fig. 8

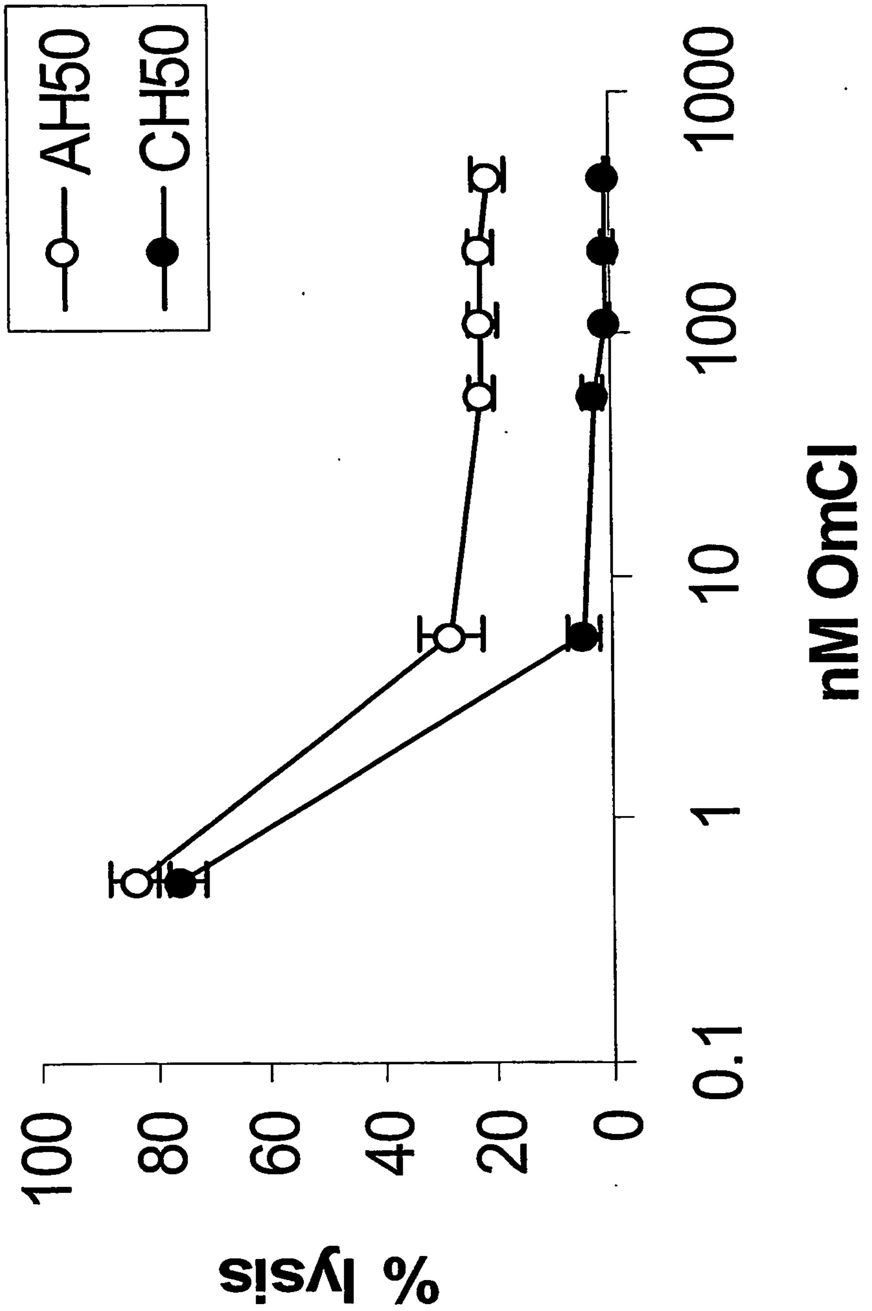


Fig. 9

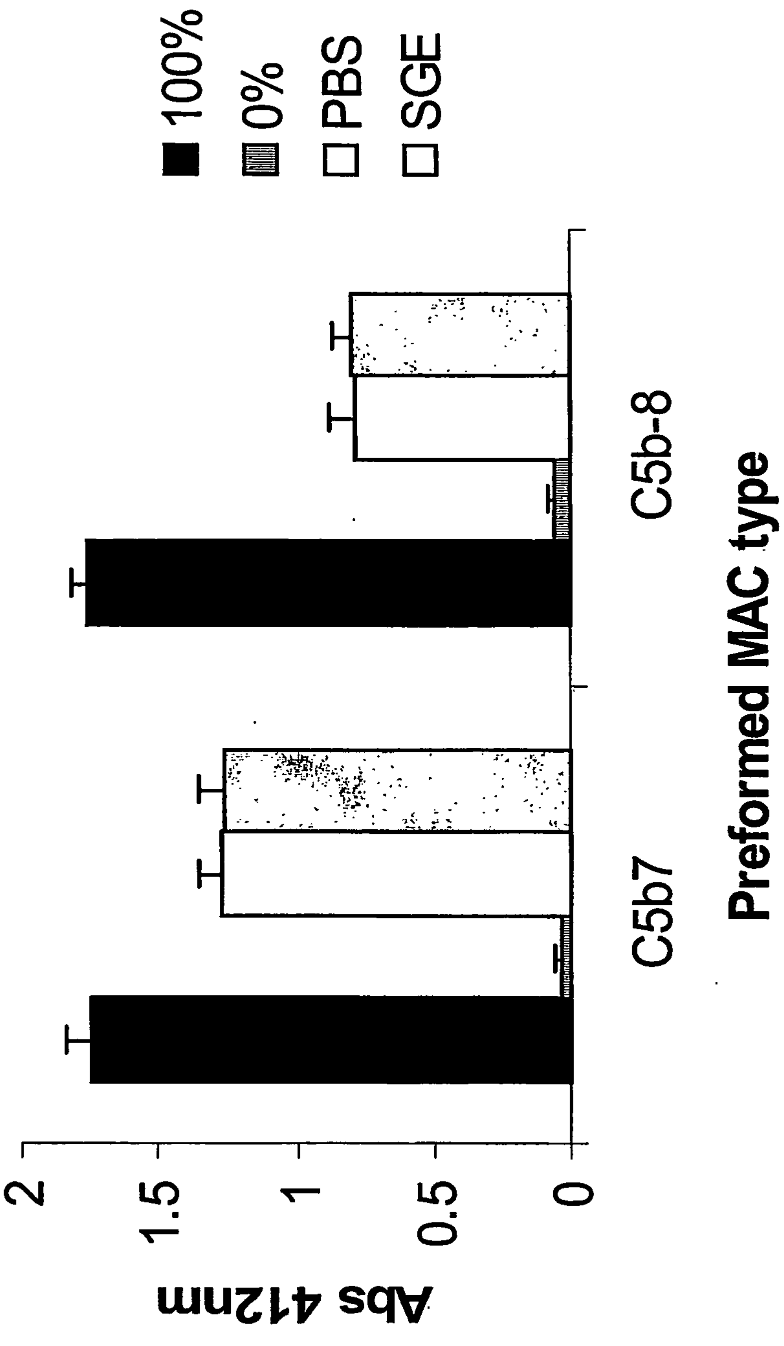


Fig. 10

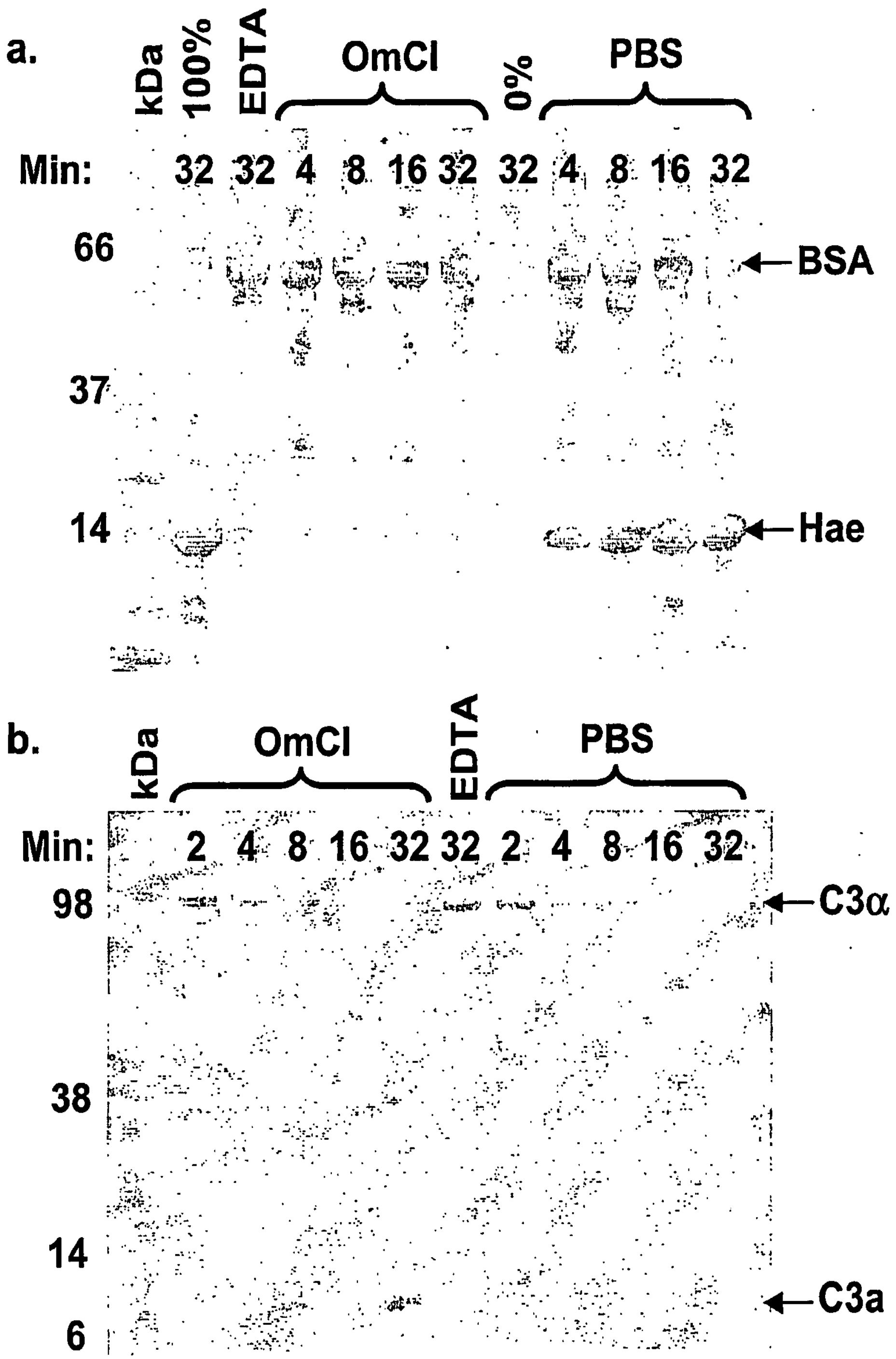

a.
kDa
100%
EDTA
OmCI
0%
PBS
Min:
32
32
4
8
16
32
32
4
8
16
32
66
37
14
BSA
Hae
b.
kDa
OmCI
EDTA
PBS
Min:
2
4
8
16
32
32
2
4
8
16
32
98
38
14
6
C3α
C3a

Fig. 11

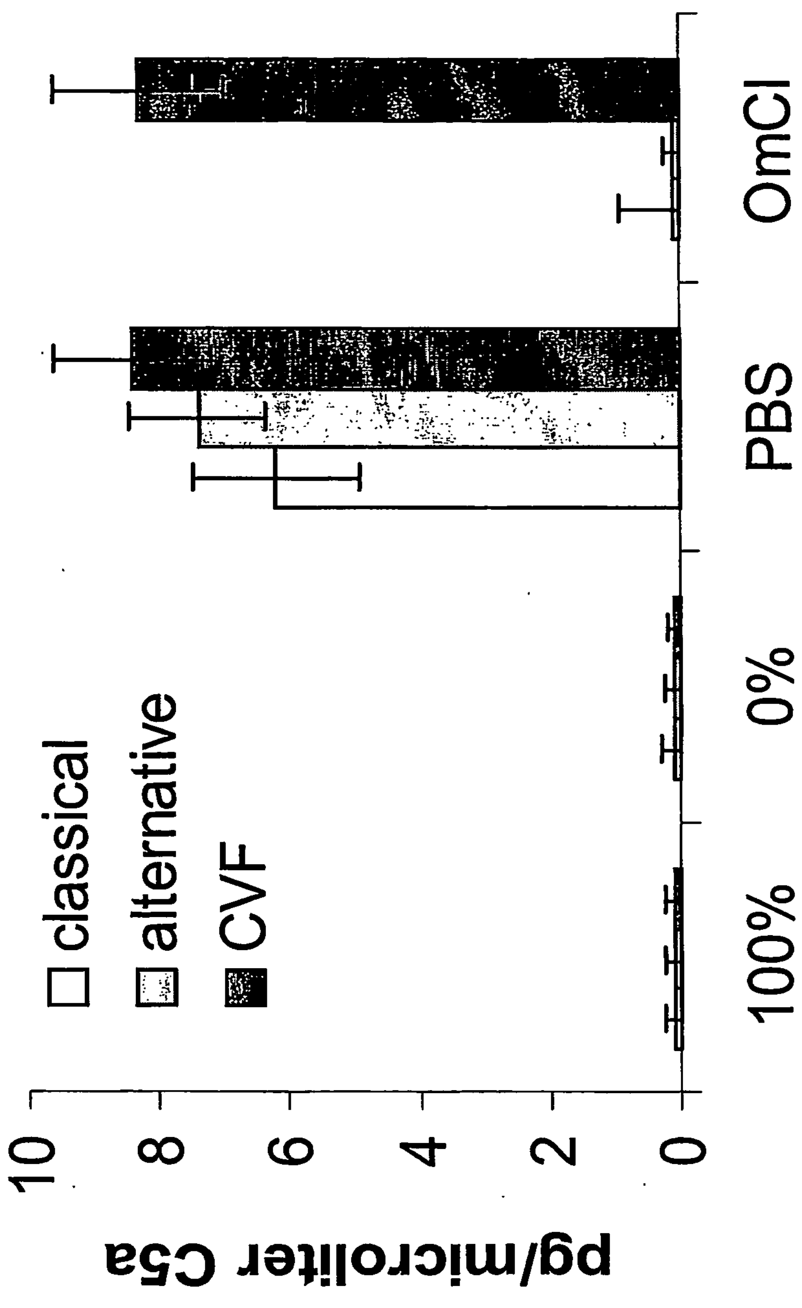

pg/microliter C5a
0
2
4
6
8
10
100%
0%
PBS
OmCI
□ classical
▨ alternative
■ CVF

Fig. 12

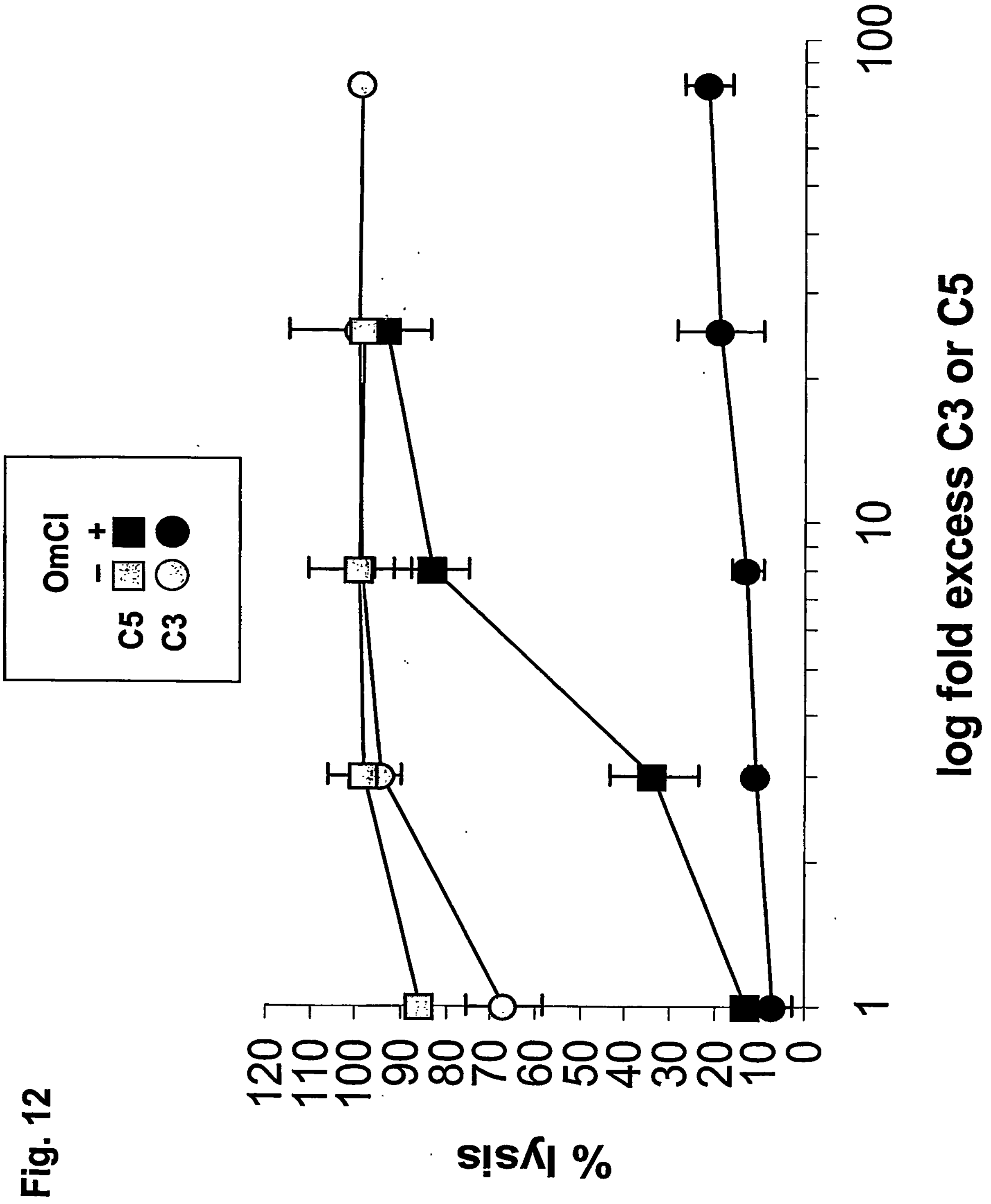
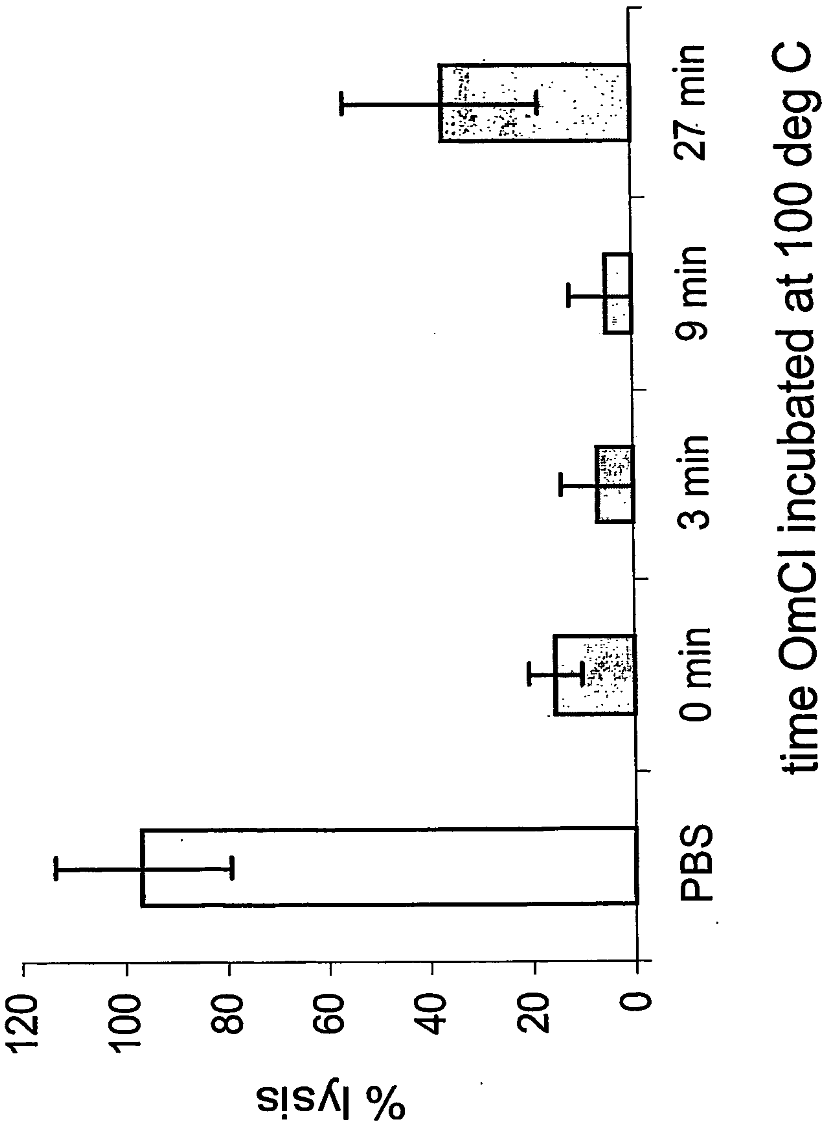

OmCI
C5 − ▨ + ■
C3 − ○ + ●
% lysis
120
110
100
90
80
70
60
50
40
30
20
10
0
1
10
100
log fold excess C3 or C5

Fig. 13


% lysis
120
100
80
60
40
20
0
PBS
0 min
3 min
9 min
27 min
time OmCI incubated at 100 deg C

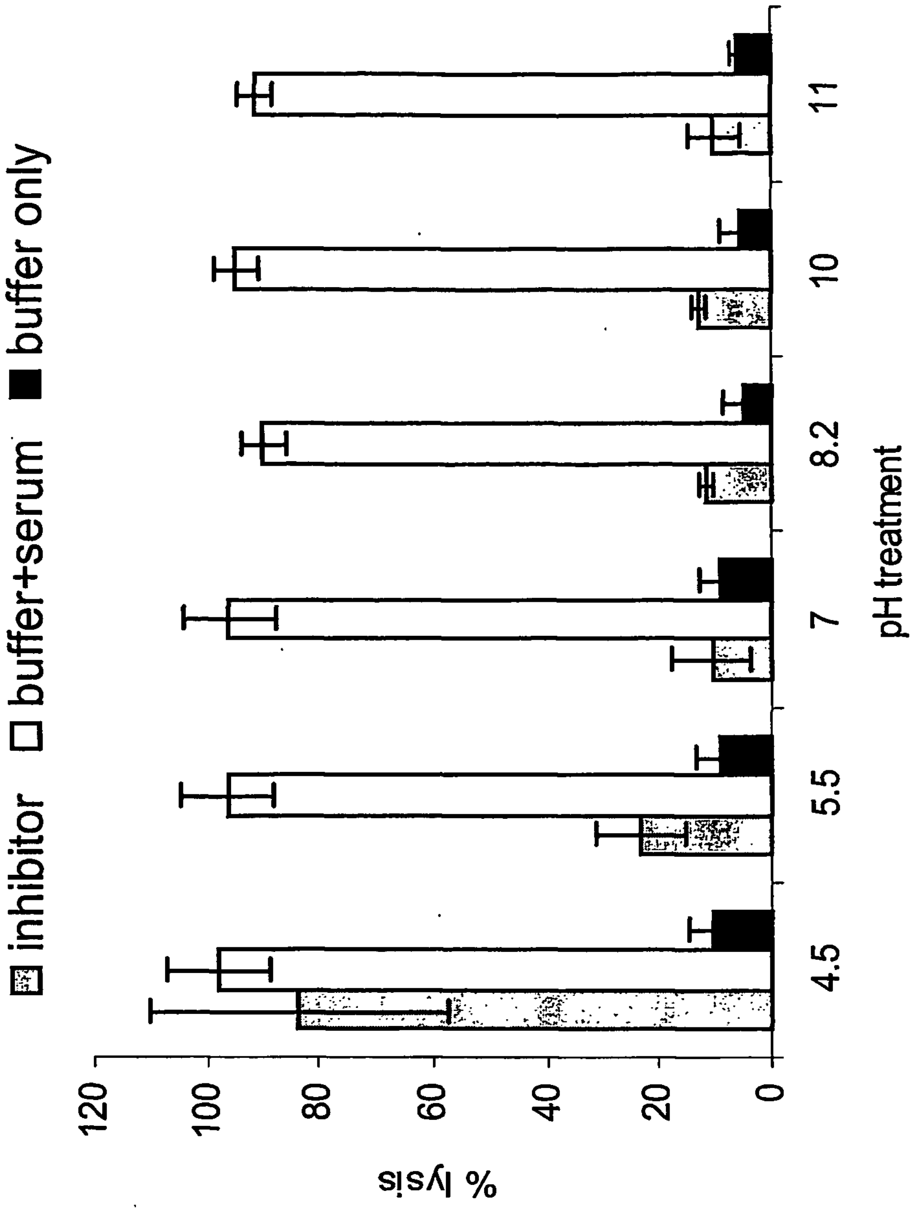

▨ inhibitor
☐ buffer+serum
■ buffer only
% lysis
120
100
80
60
40
20
0
4.5
5.5
7
8.2
10
11
pH treatment

Fig. 14

Fig. 15

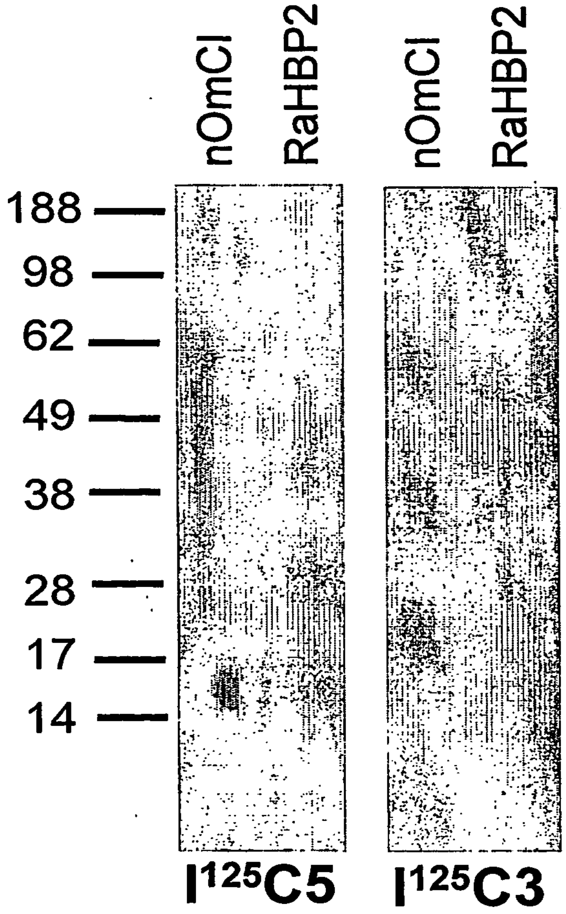
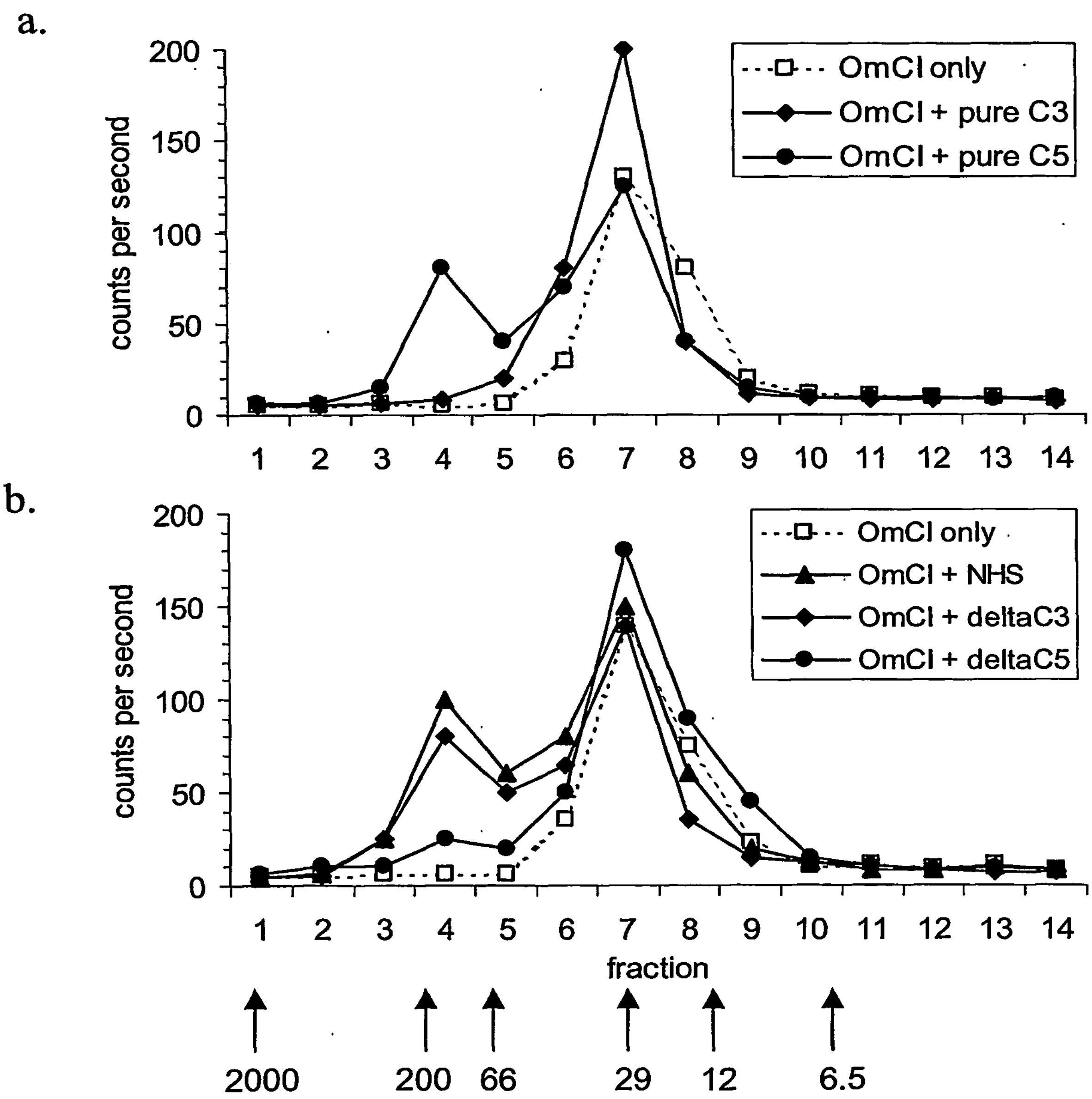

nOmCI
RaHBP2
nOmCI
RaHBP2
188
98
62
49
38
28
17
14
I125C5
I125C3

Figure 16


a.
200
150
100
50
0
counts per second
OmCI only
OmCI + pure C3
OmCI + pure C5
1
2
3
4
5
6
7
8
9
10
11
12
13
14
b.
200
150
100
50
0
counts per second
OmCI only
OmCI + NHS
OmCI + deltaC3
OmCI + deltaC5
1
2
3
4
5
6
7
8
9
10
11
12
13
14
fraction
2000
200
66
29
12
6.5